

Review of the biogeography of *Artemia* Leach, 1819 (Crustacea: Anostraca) in Algeria

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Abstract

The brine shrimp *Artemia* is a cosmopolitan Branchiopoda that lives in hypersaline water bodies where predators are rare or absent. These are concentrated in arid and semi arid regions and represented by salt Lakes and pans, salt works and the called Chotts and Sebkhass in Arabic countries. Algeria comprises the biggest desert of the world where many of these shallow endorheic depressions are formed. At least, ten among them harbor *Artemia*. The most important, in point of view

of biomass and available data, is the Chott Marouane (northeast Algeria). This paper provides main information about the sites of *Artemia*, the taxonomy, the biogeography and the ecology of known populations of this species. Based on recent phylogenetical and morphological studies, the populations recorded belong to the bisexual species *Artemia salina* and to the asexual group (2n and 4n parthenogenetical populations).

Key Words: *Artemia*, Biogeography, Algeria

Introduction

The timeline of scientific description of brine shrimp referred to known Iranian geographer, who in 982 reported its occurrence at Urmia Lake, Iran (Asem, 2008) and then in 1756 Schlösser pictured both male and female specimens from Lymington, UK. In 1758, Linnaeus assigned this species to the taxonomic classification of *Cancer salinus* (Artom, 1931). It was then renamed by Leach in 1819 to *Artemia salina* (Artom, 1931), which is currently acknowledged as the valid classification for Mediterranean bisexual populations considered conspecific with the now extinct Lymington population (Triantaphyllidis *et al.*, 1997; Baxevanis *et al.*, 2006).

The brine shrimp *Artemia* (Crustacea, Anostraca) inhabits hypersaline aquatic ecosystems worldwide, in coastal solar salterns as well as in inland salt lakes (Triantaphyllidis *et al.*, 1998; Muñoz and Pacios, 2010). It can be found in every continent, except Antarctica (Persoone and Sorgeloos, 1980; Browne and McDonald, 1982; Vanhaecke *et al.*, 1987; Van Stappen, 2002), in altitudes from as low as sea level to about 5040 m in Tibet (Yuan *et al.*, 2007) and in climatological conditions ranging from humid-subhumid to arid (Vanhaecke *et al.*, 1987). This Branchiopod has acquired adaptive mechanisms to survive and evolve in habitats with extensive and often abrupt fluctuations in salinity, UV irradiation, temperature and oxygen concentration (Persoone and Sorgeloos, 1980; Browne *et al.*, 1991; Abatzopoulos *et al.*, 2002). It is found in natural habitats with salinities from 10 g.l⁻¹ to 340 g.l⁻¹ (Sorgeloos *et al.*, 1986), but is rarely found in waters with salinity lower than 45 g.l⁻¹.

Artemia comprises a group of bisexual and parthenogenetic species, which probably diverged five to six million years ago from an ancestral form living in the Mediterranean area (Abreu-Grobois and Beardmore, 1982; Badaracco *et al.*, 1987). On the basis of criteria from morphometry and laboratory reproductive isolation, and subsequently through karyology, allozyme divergence and new molecular markers, seven bisexual species and two or three parthenogenetic forms are currently recognized in the genus *Artemia*. They all look rather similar in body shape, but show morphological traits that enable morphometric differentiation when they are cultured under

standard laboratory conditions (Hontoria and Amat, 1992a,b). Two bisexual species are endemic to the New World: *A. persimilis* Piccinelli and Prosdocimi, 1968 occurs in Argentina and Chile and *A. franciscana* Kellogg 1906 occurs in North, Central and South America. The bisexual *A. salina* (L., 1758) is found in the Mediterranean area and Africa (Asem *et al.*, 2010), *A. urmiana* Günther, 1899 in Iran, *A. sinica* Cai, 1989 in China, and *A. tibetiana* Abatzopoulos *et al.*, 1998, in Tibet. Finally, *Artemia* sp. from Kazakhstan (Pilla and Beardmore, 1994) is endemic to the Old World. Regarding the asexual populations, they are composed of di-, tri-, tetra- and pentaploid individuals or mixtures of different ploidies and are grouped under the parthenogenetic populations of *Artemia* (Triantaphyllidis *et al.*, 1998; Sun *et al.*, 1999; Gajardo *et al.*, 2002; Van Stappen, 2002; see also Abatzopoulos *et al.*, 2002; Asem *et al.*, 2010).

Artemia salina (L., 1758) is the only sexual brine shrimp species endemic to the Mediterranean Basin, and it is also found in a disjunction area in South Africa. This South African population was found to be reproductively compatible with Mediterranean isolates, but there were some significant morphological and life-history differences between both groups (Amat *et al.*, 1995). In addition, the sexual North American species *A. franciscana*, widely used as live food in aquaculture, has recently been introduced to the Mediterranean Basin and has quickly replaced native *Artemia* species in many localities (Amat *et al.*, 2005; 2007).

Despite the effort made in recent years to report on new *Artemia* populations, our knowledge of the biogeography of this anostracan is still very limited in North Africa. This paper provides an updated list of *Artemia* sites in Algeria and is based on previous publications (Sorgeloos *et al.*, 1986; Vanhaecke *et al.*, 1987; Zemmouri, 1991; Haddag, 1991; Beladjal *et al.*, 1995; Kara, 1998; Gagneur and Kara, 2001; Kara *et al.*, 2004; Samraoui *et al.*, 2006; Amarouyache, 2009; Amarouyache *et al.*, 2009a,b; Amarouyache *et al.*, 2010; Amarouyache and Kara, 2010; Derbal *et al.*, 2010; Ghomari *et al.*, 2011; Amarouyache *et al.*, 2012), as well as on personal communications (National Salt Company: ENASEL). These biog-

eographical data are discussed in connection to the most recent knowledge of morphometric and genetic data on the genus *Artemia*.

The genus *Artemia* in the Mediterranean region

In the Mediterranean region, the native species are the sexual *Artemia salina* (formerly *Artemia tunisiana*, see Belk and Brtek (1995) and the parthenogenetic populations, mainly diploid and tetraploid (Abatzopoulos *et al.*, 2002). Since the 1950s, *A. franciscana* cysts have been commercially exported worldwide from San Francisco Bay and Great Salt Lake, U.S.A., for use in aquaculture (Abatzopoulos *et al.*, 2002). In the Mediterranean area, many former saltworks in which salt production has become unprofitable are being transformed into aquaculture facilities, leading to the release of *A. franciscana* into hatcheries and its escape into habitats where native *Artemia* populations occur (Amat *et al.*, 2005).

Artemia cysts from numerous saltworks in the West Mediterranean were collected, and their species composition determined (Amat *et al.*, 2005). The American species was first detected in 1981 in the Portuguese Algarve and was the only species recorded in this country, where it is found in areas both with and without aquaculture. It has since spread to all the Iberian sites considered to be most important for shorebirds along the East Atlantic Flyway (Boyd and Pirot, 1989; Amat *et al.*, 2005). It is also found in Cadiz Bay (Spain), two French sites, and one in Morocco (Sebkha Bou Areg) (Amat *et al.*, 1987, 2005; Thiéry and Robert, 1992; Green *et al.*, 2005). Mura *et al.* (2006) revealed its presence in Margherita di Savoia Salterns in Italy and more recently, Ben Naceur *et al.* (2010b) reported its presence and characterized it in Tunisia (Sebkha Halk El Menzel).

Artemia biodiversity in the Mediterranean Basin is probably unique in comparison to other parts of the organism's natural distribution (Mura *et al.*, 2006). In this region, varieties of parthenogens are found in coexistence or have possibly evolved sympatrically with the native sexual *A. salina* (Amat, 1983; Lenz and Browne, 1991; Amat *et al.*, 1995). Throughout southern Europe and North Africa, this pattern of

interaction within and between parthenogenetic types and bisexuals has provided rare opportunities for investigating the partitioning of genetic variation and the differentiation of life-history traits and ecological attributes (Browne *et al.*, 1991; Zhang and King, 1992, 1993; Barata *et al.*, 1996a,b; Abatzopoulos *et al.*, 2002; Baxevanis *et al.*, 2006).

Just recently, Muñoz *et al.* (2008) used partial sequences of the mitochondrial cytochrome *c* Oxidase Subunit 1 (COI or *cox1*) gene to investigate the genetic diversity and phylogeography of *A. salina* over its known distribution range (Mediterranean Basin and South Africa) and to assess the extent of local endemism, the degree of population structure and the potential impact of traditional human saltpan management on this species. The authors conclude that *A. salina* populations hold substantial genetic diversity, with a strong phylogeographical structure and high regional endemism in Mediterranean Basin, Iberian Peninsula, North Africa and on Italian Islands (Sardinia and Sicily). Their analyses point to an episode of population expansion in the area, possibly during the early Pleistocene, followed by more localized expansions through recolonization routes. These expansions sometimes involved long-distance colonization or isolation and genetic fragmentation. The signature of correlation between genetic and geographical distance could instead reflect a pattern of sequential colonization due to an ancient founder effect. The authors indicate that the *A. salina* population from South Africa is highly differentiated genetically from the Mediterranean populations and might deserve specific status. Their results also indicate that traditional human saltpan management used to be compatible with the maintenance of population genetic diversity in this organism. However, recent changes of saltpan use, such as aquaculture, which promotes the introduction of the alien species *A. franciscana*, as well as habitat loss, pose serious threats for the genetic diversity of *A. salina* populations in the Mediterranean area.

In the Mediterranean region, *Artemia* species have been investigated and characterized only in the European sites (Triantaphyllidis *et al.*, 1993) while in the south of the Mediterranean Basin, where promising *Artemia* sites exist as in Algeria (Haddag,

1991; Kara *et al.*, 2004; Samraoui *et al.*, 2006; Amarouyache, 2009; Amarouyache *et al.*, 2009a,b, 2010; Amarouyache and Kara 2010; Derbal *et al.*, 2010; Ghomari *et al.*, 2011; Amarouyache *et al.*, 2012), in Tunisia (Van Ballaer *et al.*, 1987; Aloui, 2003; Ben Naceur *et al.*, 2008, 2009, 2010a,b, 2011), in Morocco (Sadkaoui, 2000), in Egypt (El Bermawi, 2003, 2004) and in Libya (El Magsodi *et al.*, 2005), only few data about biology, nutritional quality and reproductive characteristics of *Artemia* are available. Until recently, biogeographical and biodiversity studies dealing with the distribution of *Artemia* species in North Africa have been limited to the identification of populations belonging to the bisexual *A. salina*, while the presence of parthenogenetic strains has been scarcely known (Amat, 1983; El-Bermawi *et al.*, 2004).

Biogeography of *Artemia* in Algeria

-Geography and climate of Algeria

Algeria is the largest African country (about 2.4 10⁶ km²). It ranges from the Mediterranean coast in the North down to below the 20th parallel in the South. So most of its territory is occupied by the well-known Sahara desert. Over 85% of the country's area corresponds to desert or sub-desert zones. Mainly lying under arid or semi-arid climates, surface water resources are scarce, irregular and extremely localized. Average annual rainfall is approximately 70 mm but it ranges from near 0 mm in the Sahara desert up to 1500 mm in the northeastern coastal area. Even in this relatively wet region, annual rainfall variability lies between 20 and 50% (Bensaad, 1988). One can very easily distinguish two climatic zones:

- Northern Algeria, or Tell, with a typical Mediterranean climate. Here the hydrographic network is mainly composed of temporary watercourses, called wadis, and reservoirs built for drinking water supplies and irrigation. Wadis are streams characterized by a very low base flow and some of them can become dry for several months. But at the opposite, they are exposed to severe flash floods that scour their beds and their banks, often destroying bridges and changing bed location.
- Algeria south of the Tell Atlas Mountains that overlook the High Plains and the desert, has an arid

climate with often far less than 300 mm rainfall per year. There, all the basins are endorheic and surface water is of three types: (1) ephemeral watercourses following fossil valleys indicated by lines of oases (this illustrating the uses of underflow for millennia), (2) temporary freshwater ponds called Dayas and Gueltas, mostly small in area and (3) perennial or temporary saline lakes called Chotts or Sebkhass.

-The occurrence of *Artemia* in saline lakes in Algeria

In Algeria, brine shrimp are especially associated with Sebkhass and Chotts, terms of Arabic origin. They are shallow depressions filled with saline waters. Filling is irregular over time and space (Morgan, 1982). Some of them were lakes during the damper periods of late Quaternary. According to the terminology accepted by North African geomorphologists, a Sebkhass is a closed inland basin with impermeable bottom, which accumulates soluble and insoluble products provided by water inflows from surroundings, large hydrological areas. It can be filled up and flooded during short heavy rain periods, but becomes dry, shortly afterwards because of evaporation. A Chott usually does not become completely dry, as it relies on underground water inflow. The biggest Chotts in Algeria are located on the high plains between the two Atlas ranges, and from west to east are: Chott Ech-Chergui, Chott El Hodna, Chott Melghir.

The presence of *Artemia* in hyperhaline lakes in Algeria is reported for the first time by Blanchard and Richard (1890), then in 1928 by Gauthier (Gauthier, 1928) and in 1931 by Seurat (Seurat, 1931). While searching for sites likely to shelter the Branchiopod Crustacea *Artemia* species, Sorgeloos *et al.* (1986) listed nine hyperhaline water bodies in Algeria (Tab. 1), some of which are exploited for salt production (Ez-zemoul, Djeloud, Guergour El-Amri, Marouane, Bethioua, Sidi Bouziane). Zemmouri (1991) mentioned five sites: Sebkhass Arzew, Sebkhass N'Zouri, Sebkhass Sidi Bouziane, Chott Marouane, Mellaha Guergour El-Amri. Nevertheless, the comparison of geographical coordinates shows that 4 of them were already cited but with different names Sebkhass Arzew/ Bethioua saltworks or Sebkhass, Sebkhass N'zouri/

Table 1: *Artemia* sites in Algeria (B: bisexual population; P: parthenogenetic population; 2n, 4n: indicates the ploidy level of a parthenogenetic population (2: diploid, 4: tetraploid))

Locality	Geographical Coordinates	Province	Area (ha)	Depth (m)	Mode of Reproduction	Species	Ref.
Sebkhet Oran	35°32'N-00°48'W	Oran	43,000		P(2n)		1,14
Dayet Morseli	35°30'N-00°46'W	Oran	150		?	?	1
Gharabas Lake	35°35'N-00°25'W	Oran	< 1		?	?	1
Chott Ouargla	31°57'N-05°20'E	Ouargla	6,853		?	?	1
Chegga Oase	34°29'N-05°53'E	El Oued	1?		?	?	1
Chott Touggourt	33°06'N-06°07'E	Touggourt	< 1		?	?	1,4
Chott Marouane (Djeloud)	34°03'N-06°20'E	El Oued	36,000	40 (max.)	B	<i>A. salina</i>	1,2,7,8,9,10
Sebkha Ez-Zemoul	35°53'N-06°33'E	Oum El Bouaghi	6,100	0.6	B, P(2n)	<i>A. salina</i>	1,2,5,8,10,11,12,14
Arzew Saltern (Bethioua)	35°41'N-00°17'W	Arzew	2,900		P(4n), P(2n)		2,3,8,14
Mellaha Guergour El-Amri	35°59'N-05°15'E	Sétif	< 1		?	?	2
Sebkha N'zouri (Ez-Zemoul)	35°53'N-06°27'E	Oum El Bouaghi	6,100		?	?	5,6
Garaet El Tarf	35°42'N-07°07'E	Oum El Bouaghi	33,460	1 (max.)	B	<i>A. salina</i>	5,6,13,14
Chott Melghir	34°10'N-06°17'E	Biskra	48,000		B, P(2n)	<i>A. salina</i>	8,14
Relizane Sebkha	35°50'N-00°39'W	Sidi Bouziane	1,740		B, P(4n), P(2n)	<i>A. salina</i>	2,14
El-Bahira Lake	35°50'N- 05°15'E	Rosfa Sétif	10		P		1,14, 15
Goléa Salt Lake	30°28'N-02°55'E	Ghardaia	18,947	2 (max.)	P(4n)		14

(1) Sorgeloos *et al.* (1986), (2) Zemmouri (1991), (3) Haddag (1991), (4) Beladjal *et al.* (1995), (5) Kara (1998), (6) Gagneur and Kara (2001), (7) Kara *et al.* (2004), (8) Samraoui *et al.* (2006), (9) Amarouyache *et al.* (2009a), (10) Amarouyache and Kara (2010), (11) Amarouyache *et al.* (2010), (12) Amarouyache *et al.* (2012), (13) Muñoz *et al.* (2008), (14) Ghomari *et al.* (2011), (15) Derbal *et al.*, (2010).

Sebkha Ez-Zemoul, Sebkha Sidi Bouziane / Djemaa Lake and Chott Marouane / Chott Djeloud (Sorgeloos *et al.*, 1986). With the same aim, four other haline sites have been surveyed by Kara (1998) (El Ghaba, Ed-dar, Melha, Magtaa) in the neighbourhood of Chott Marouane called Chott Djeloud by Morgan (1982) and Sorgeloos *et al.* (1986). These are devoid of *Artemia* (Kara, 1998), as well as Sebkha Djendli (Batna district) where *Branchinecta media* Schman-kewitsch 1873 was identified as *Artemia salina* by Blanchard and Richard (1890) (Samraoui *et al.*, 2006; Amarouyache, 2009). Ghomari *et al.* (2011) used the name of Sebkha Sétif instead of El-Bahira Lake (Rosfa district) which is the original name used by the local population. Table 1 provides a list of all known *Artemia* sites in Algeria. The geographical coordinates and areas are given (Anonymous 1998) and, when available, information about the depth and the mode of reproduction (parthenogenetic or bisexual), the ploidy level of the parthenogenetic populations and

the species designation.

-Biodiversity in the genus *Artemia*

Despite the abundant and wide distribution of hypersaline areas in Algeria, very little is known about the biodiversity of *Artemia* populations inhabiting them. Amarouyache (2009), using scanning microscopy was based on the morphology of males (frontal knob morphology and ornamentation as well as the basal part of the penis) to attach the brine shrimp of Chott Marouane and Sebkha Ez-Zemoul to the species *A. salina* (Fig. 1). Ghomari *et al.* (2011) chose morphometry as a tool to identify the different species and strains that are present in eight sites in Algeria (Tab. 1). The results obtained with this method have been confirmed by using Mediterranean reference species and populations that were taxonomically described and studied in multidisciplinary approaches, including molecular biology studies (Mura *et al.*, 2005, 2006; Muñoz *et al.*, 2008, 2010). All populations

studied, either cultured under laboratory conditions after hatching original cysts, or observed from preserved samples collected in the wild, showed more than one species or strain (Fig. 2). The only exceptions were the samples obtained from the inland Goléa Salt Lake, the Garaet El Tarf and the Oran Sebkhha, which always produced a tetraploid parthenogenetic *Artemia*, a bisexual *A. salina* and a diploid strain, respectively. *A. salina* and the diploid parthenogenetic populations co-occur in all Sebkhhas

and in the Chott Melghir, with the exception of Garaet El Tarf where only *A. salina* appeared and of Oran Sebkhha where only the diploid parthenogenetic strain is present. The tetraploid parthenogenetic population is present in the Arzew Saltern, Relizane and Setif Sebkhhas, but is exclusive for the Goléa Salt Lake. Thus, it is possible to accept that the biodiversity of the Algerian *Artemia* populations is congruent with the biodiversity found for the rest of the western Mediterranean countries (Amat *et al.*, 1995).

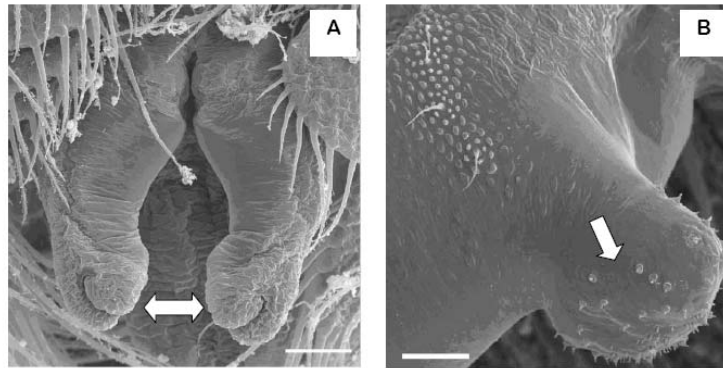


Fig. 1: SEM micrographs of *Artemia salina* male (Chott Marouane). (A) penis devoid of spine-like (x 50). (B) frontal knob with subconical shape (x 200) (Amarouyache, 2009)

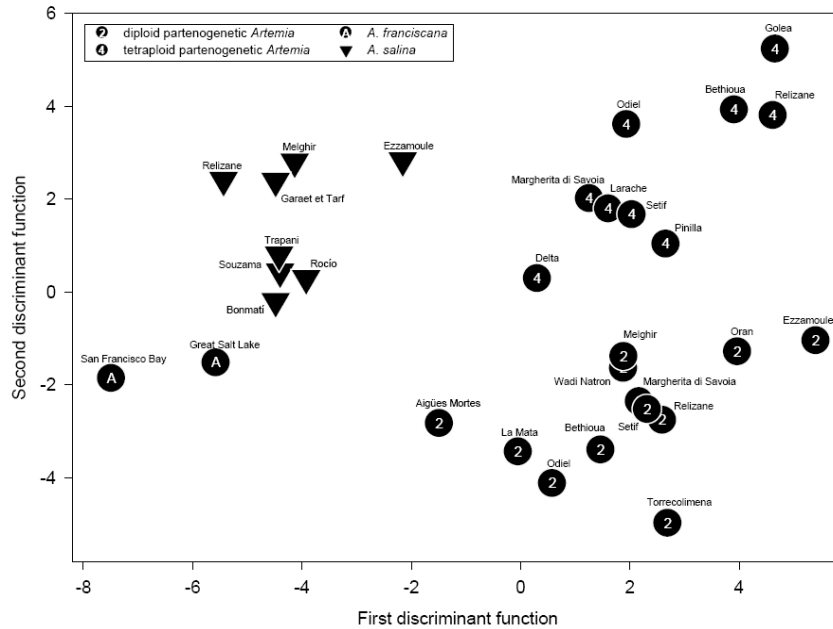


Fig. 2: Population centroids for the first functions of the discriminant analysis on the morphology of females from different Algerian *Artemia* populations, compared to several related populations from the Mediterranean area (Ghomari *et al.*, 2011)

Concerning *A. franciscana*, it is possible to expect that viable cysts of this species could eventually reach Algerian Chotts and Sebkhass consistently with a man-mediated introduction or dispersion via shorebirds (Green *et al.*, 2005).

-Ecology and biology

The most important studies that describe the ecology and the biology of *Artemia* in Algeria concern the Chott Marouane (Kara *et al.*, 2004; Amarouayache, 2009; Amarouayache *et al.*, 2009a,b; Amarouayache and Kara, 2010) and the Sebkhass Ez-Zemoul (Amarouayache, 2009; Amarouayache *et al.*, 2010, 2012). The approached aspects are mainly the biometrics and the population dynamics. During the two last decades, seasonal monitoring of Sebkhass Arzew, Chott Marouane, Sebkhass Ez-Zemoul, El-Bahira Lake showed that both bisexual (Arzew, Marouane and Ez-Zemoul) and parthenogenetic (El-Bahira) species develop only in winter and spring, even when the pools are full of water in summer and autumn (Haddag, 1991; Amarouayache *et al.*, 2009a, 2010; Derbal *et al.*, 2010). The maximal recorded salinity in the wild is 250 g.l⁻¹ for Sebkhass Ez-Zemoul and Sebkhass Arzew and 360 g.l⁻¹ for Chott Marouane (Haddag, 1991; Amarouayache *et al.*, 2009a; 2010). In the Sebkhass Ez-Zemoul where salinity decreases until 30 g.l⁻¹, *Artemia* co-occurs with other crustaceans as *Phallocryptus spinosa*, *Cletocamptus retrogressus*, *Moina salina* and *Heterocypris* sp. (Amarouayache *et al.*, 2012). The highest densities of *Artemia* are reached in March/April with 56 ind.l⁻¹ in Sebkhass Ez-Zemoul and 107 ind.l⁻¹ in El-Bahira Lake, which is considered as very high by Lavens *et al.* (1986). Even if parthenogenetic and bisexual species co-habit together according to Ghomari *et al.* (2011), the sex-ratio is always in favor of males in Chott Marouane, Sebkhass Arzew and Sebkhass Ez-Zemoul (Haddag, 1991; Amarouayache *et al.*, 2009a) and increases with salinity in the latter site until 32 males/female (Amarouayache *et al.*, 2010). The populations cited can reproduce as well by oviparity as by ovoviviparity, but the first mode is always dominant (90%). Females of Chott Marouane are the less fertile and produce 10 offspring/brood against 60 offspring/brood in Sebkhass Ez-Zemoul, probably because of the high salinities of

the medium ranging between 250 and 360 g.l⁻¹ (Amarouayache *et al.*, 2009a; 2010). In situations of strong salinity, the females must save their energy reserves to resist by producing fewer cysts (Barata *et al.*, 1995). The total biomass of cysts produced in Chott Marouane (36,000 ha) is estimated, by direct observations, at 7.6 t (Amarouayache and Kara, 2010). Hydrated cysts and freshly hatched nauplii of Chott Marouane are the smallest among Algerian strains: 236.5 and 428.7 µm, respectively, vs 247.86 and 460.43 µm for Sebkhass Ez-Zemoul, 239.1 and 460 µm for Sebkhass Arzew. The biggest sizes are observed in the parthenogenetic strain of El-Bahira with 277.26 µm for cysts and 549.12 µm for nauplii. This corroborate with the conclusions of Amat (1980) for the Mediterranean populations who admits that parthenogenetic cysts, nauplii and adults are bigger than bisexual ones.

Artemia is intermediate host of 14 cestode species (Georgiev *et al.*, 2005; Vasileva *et al.*, 2009). Parasitism by Flamingo parasite *Flamingolepis liguloides* (cestoda) has been reported in some Mediterranean populations: the French Camargue (Thiéry *et al.*, 1990; Robert and Gabrion, 1991), Bonmati Salterns in Spain (Amat *et al.*, 1991), salt works of Sardinia (Mura 1995), Andalucia (Georgiev *et al.*, 2005, 2007), and for the first time in North Africa by Amarouayache *et al.*, (2009b). Cysticercoïdes of *F. liguloides* infect the juveniles and the adults of *Artemia* of Chott Marouane in winter and Sebkhass Ez-Zemoul in spring with a prevalence which attains 33 %. They have been found inside the thorax, the abdomen or the ovisac. The parasite has negative effect on reproduction for these populations, reducing fecundity or causing the castration of the females, which is the most dramatic effect known in the genus of *Artemia* (Amat *et al.*, 1991).

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